

Supporting Information

Culture conditions and isolation of compound **8**

Dickeya zae strain DZ1 was grown at 28°C on yeast extract broth (YEB) agar, which contains (per liter) 10 g Bacto tryptone, 5 g yeast extract, 5 g sucrose, 5 g NaCl, 15g agar and 1mM MgSO₄.7H₂O (pH 7.0) for 24 h. Single colonies were used to inoculate YEB broth (50 ml in a 250 ml conical flask) and incubated at 28°C (200 rpm) for overnight to prepare start culture. The minimal medium (K₂HPO₄ 10.5 g/l, KH₂PO₄ 4.5 g/l, (NH₄)₂SO₄ 2 g/l, mannitol 2 g/l, MgSO₄.7H₂O 0.2 g/l, FeSO₄ 5 mg/l, CaCl₂ 10 mg/l and MnCl₂ 2 mg/l; pH 7.0) was inoculated with start culture (0.2% v/v) and grown at 28°C for 24 h. Cells were removed by centrifugation at 13,000 rpm, 15°C for 15 min. The supernatant was adjusted to pH 10 with 5 M NaOH. After basification, the supernatant was concentrated by 10 folders by rotary evaporation and extracted by organic solvent (n-butanol : ethyl acetate = 2:1) twice (3 vol). The extract was subjected to gel filtration chromatography (GFC) on Sephadex LH-20 after remove organic solvent by rotary evaporation. MeOH was used to elute GFC. Fractions with antibiotic activity were pooled following bioassay results. Further purification was carried on by gradient HPLC on C18 reverse phase column (Phenomenex Luna 5 μ C₁₈ 250 × 4.60 mm) eluted with MeOH in H₂O (0-5 min 5%, 5-50 min 5%-75%, 50-51 min 75%-95%, 51-55 min 95%, 55-56 min 95%-5% and 56-60 min 5%; 1ml/min) and the peaks were monitored by PDA detector (Waters PDA 996) with λ = 210 nm. Bioassay data showed that retention time of zeamine **8** is 31.0 min.

Bioassay was carried on according to published protocols.^{1,2} The minimum inhibition concentration (MIC) data are summarized in Table-1.

Table 1. MIC of Zeamine **8** on Gram-positive and Gram-negative bacterial strains^a

Bacterial strains	Zeamine 8 ($\mu\text{g/ml}$)	Ampicillin ($\mu\text{g/ml}$)
<i>Staphylococcus aureus</i> ATCC 25531	0.3	15.6
<i>Staphylococcus aureus</i> BBA-41 ^b	0.3	>500
<i>Bacillus cereus</i> XJ8	3.0	100
<i>Pseudomonas aeruginosa</i> PAO1	5.0	400
<i>Pseudomonas aeruginosa</i> PA14	6.0	800
<i>Burkholderia cepacia</i> H111	50	>800
<i>Burkholderia cepacia</i> J2315	25	>800
<i>Enterobacter aerogenes</i> ATCC 13048	3.0	>800
<i>Klbsiella pneumonia</i>	6.0	>800
<i>Salmonella enterica</i>	3.0	12.5
<i>Escherichia coli</i> CFT073	3.0	25.0
<i>Escherichia coli</i> DH5 α	0.5	25.0
<i>Aeromonas hydrophila</i>	10.0	125
<i>Mycobacterium smegmatics</i>	3.0	400

a: Ampicillin was used as a comparison control. b: methicillin resistant *Staphylococcus aureus* ³

Reference:

1. M. B. B. M. Hussain, H.-B. Zhang, J.-L. Xu, Q. Liu, Z. Jiang and L.-H. Zhang, *J. Bacteriol.*, 2008, **190**, 1045-1053.
2. National Committee for Clinical Laboratory Standards. (2000). *Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria that Grow Aerobically—Fifth Edition: Approved Standard M7-A5*. NCCLS, Wayne, PA.
3. H. Sato and J. B. Feix, *Antimicrob. Agents Chemother.* 2008, **52**, 4463-4465.

Supplementary Figures

xj-lh9 rp31RAW #8-25 RT: 0.23-0.71 AV: 18 NL: 6.60E5
T: + p Full ms [50.00-2000.00]

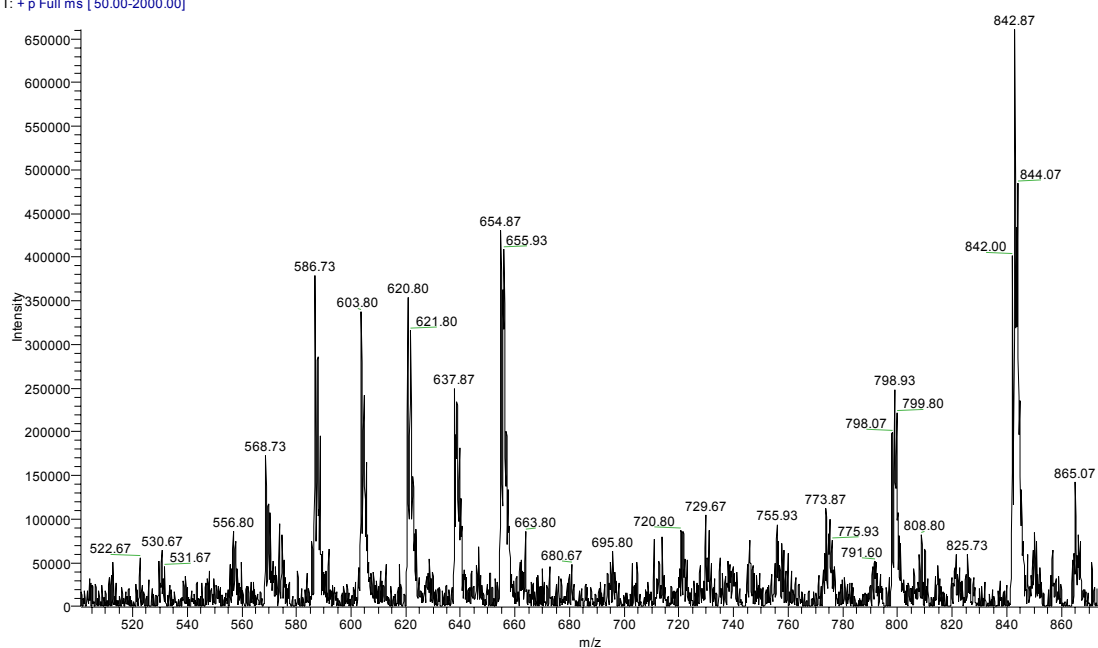


Figure 1. ESI MS analysis of zeamine **8**.

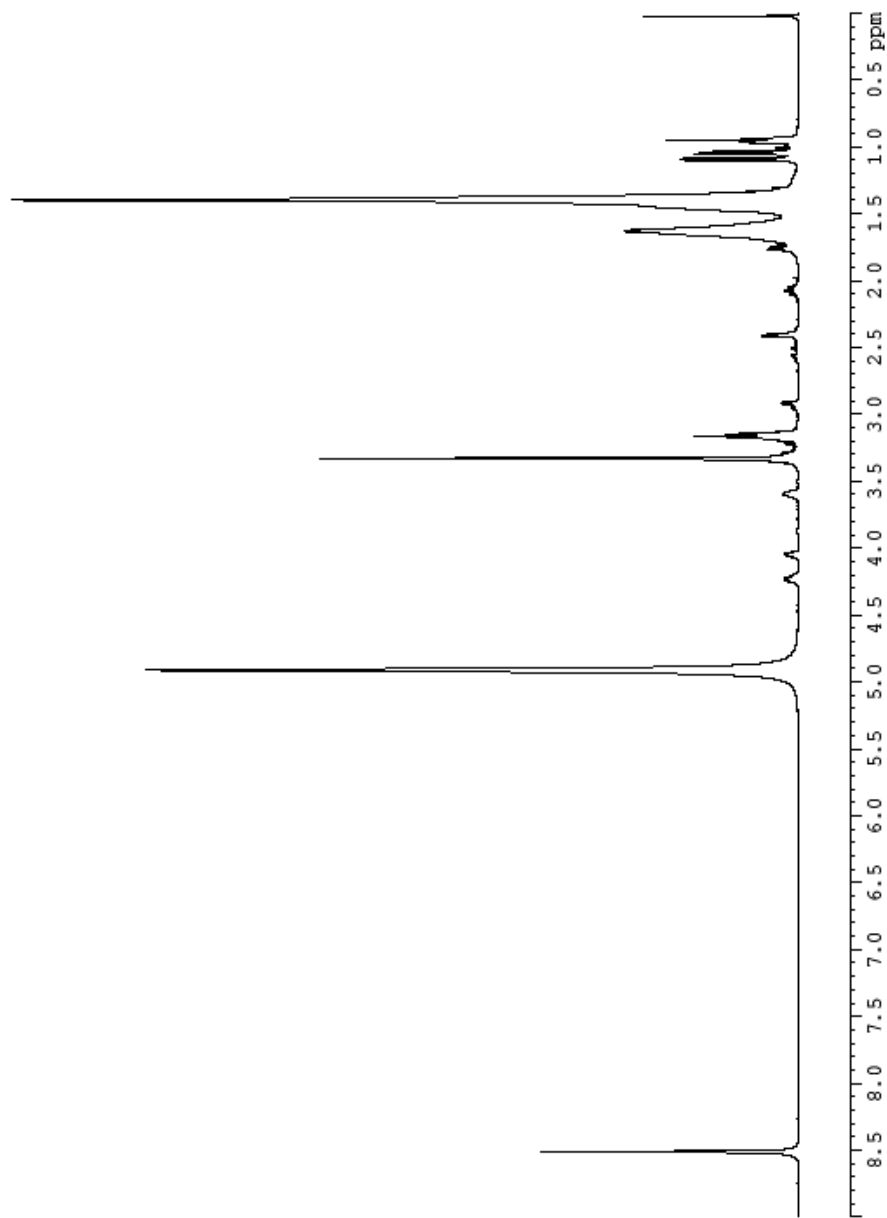


Figure 2. ¹H NMR spectrum of compound **8**

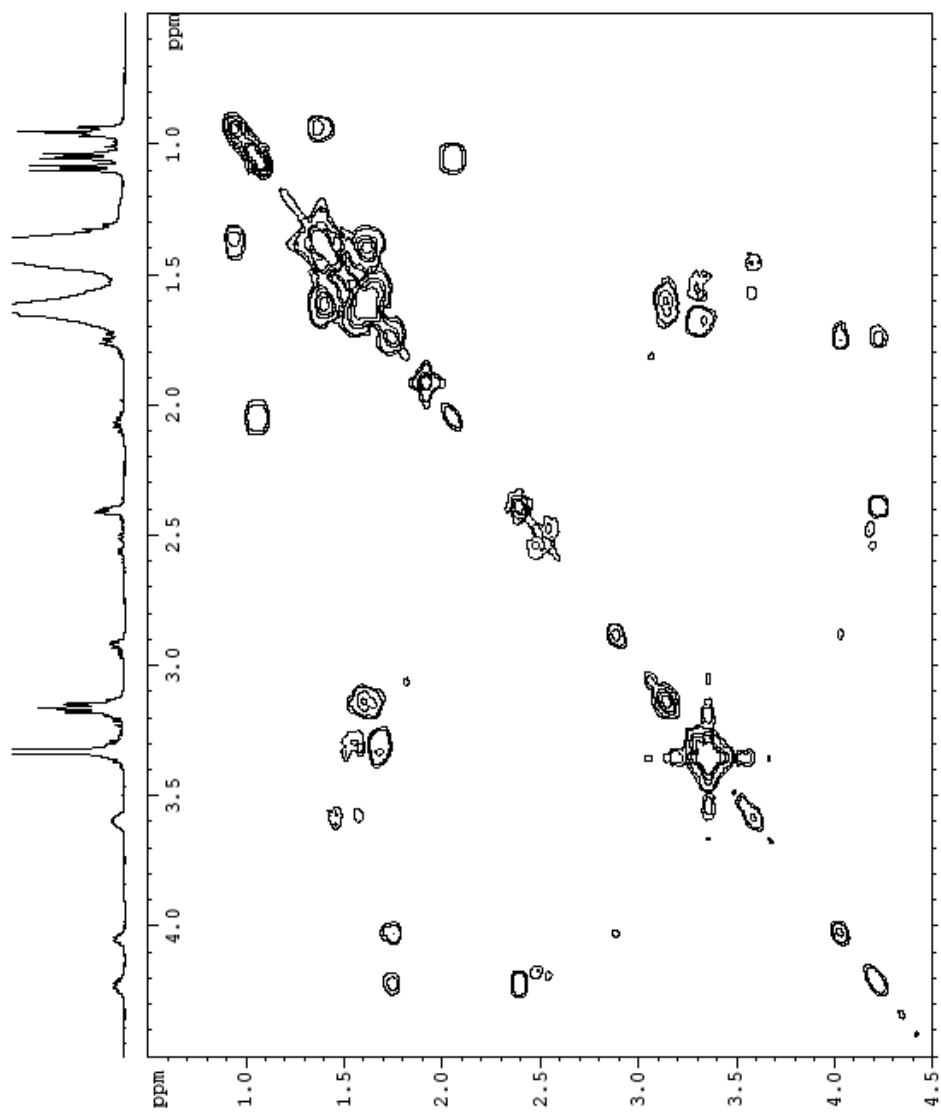


Figure 3. ¹H COSY NMR spectrum of compound **8**

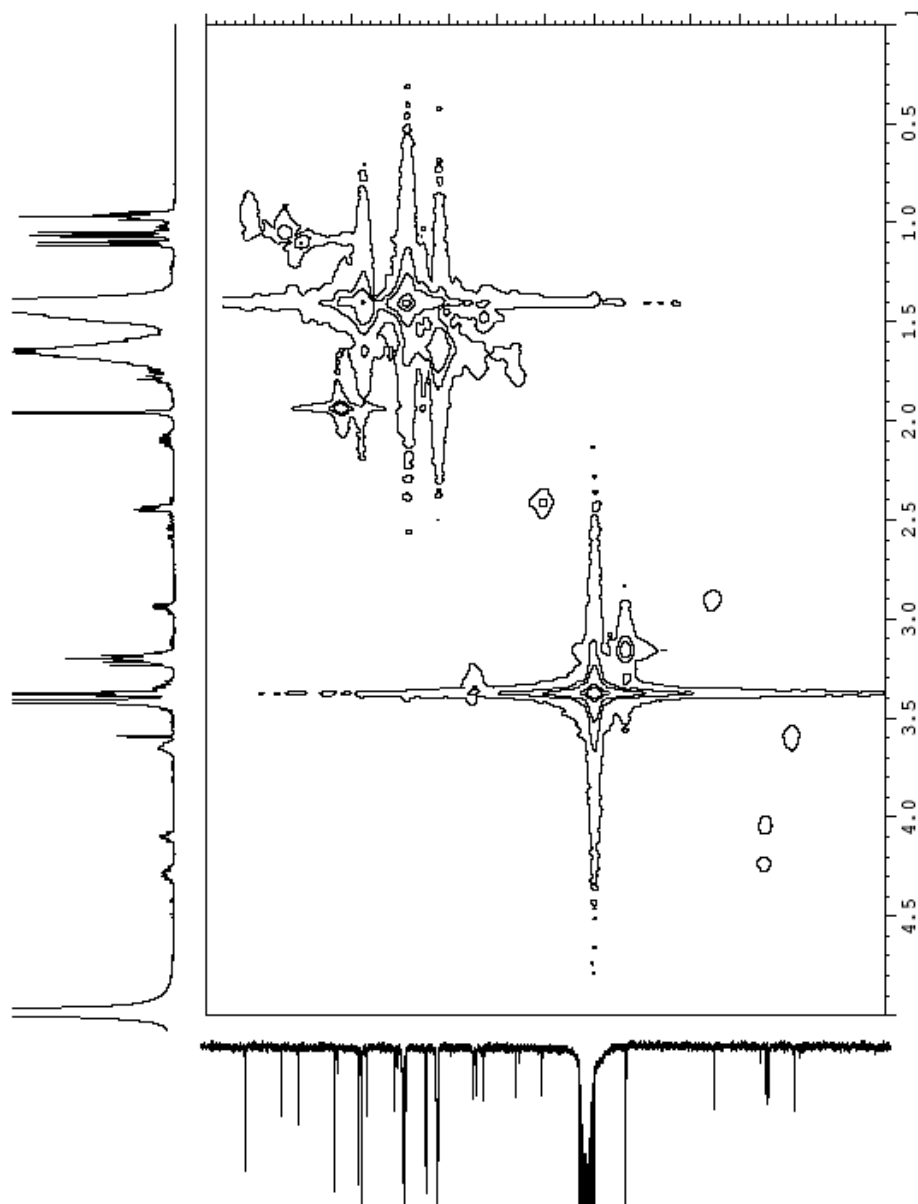


Figure 4. ^1H , ^{13}C HMQC NMR spectrum of compound **8**

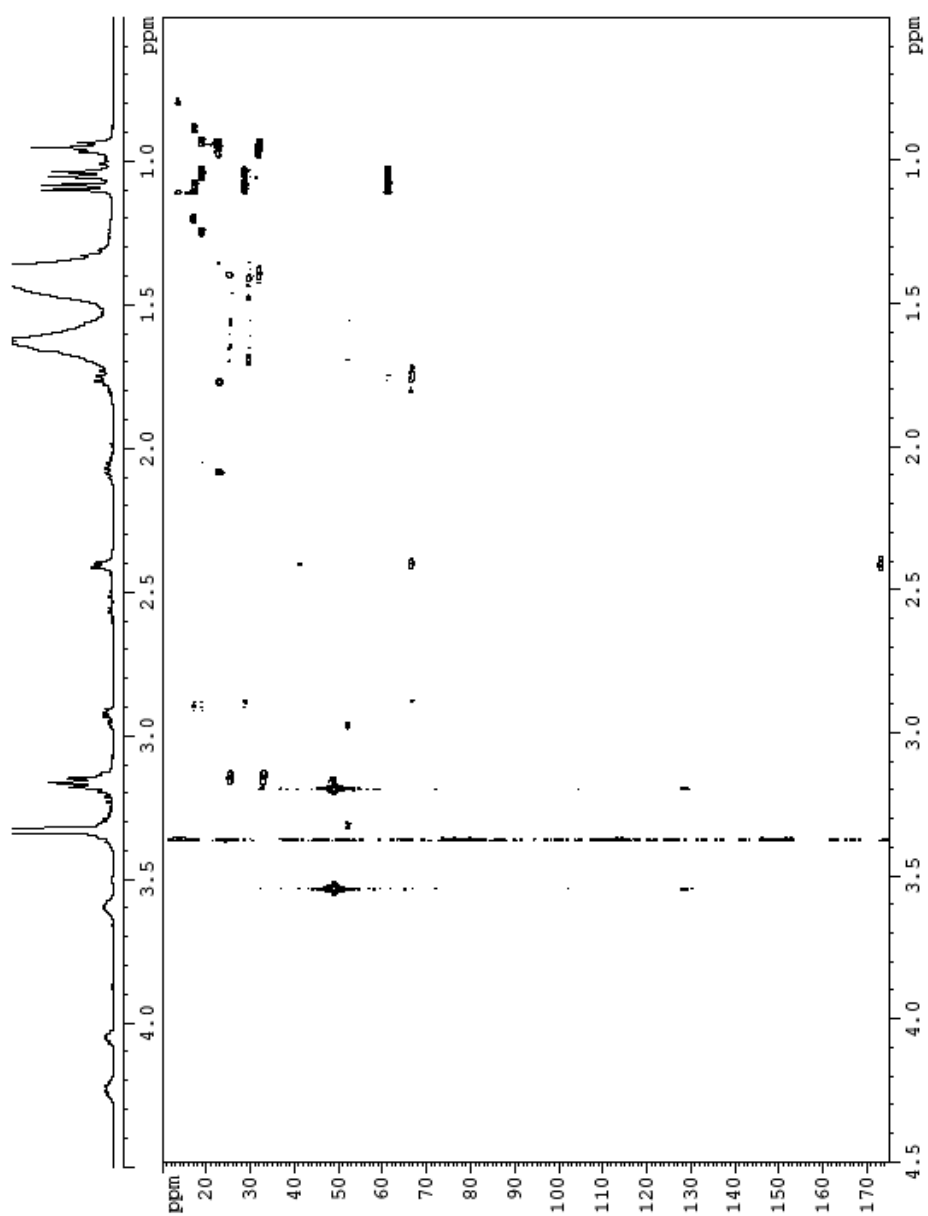
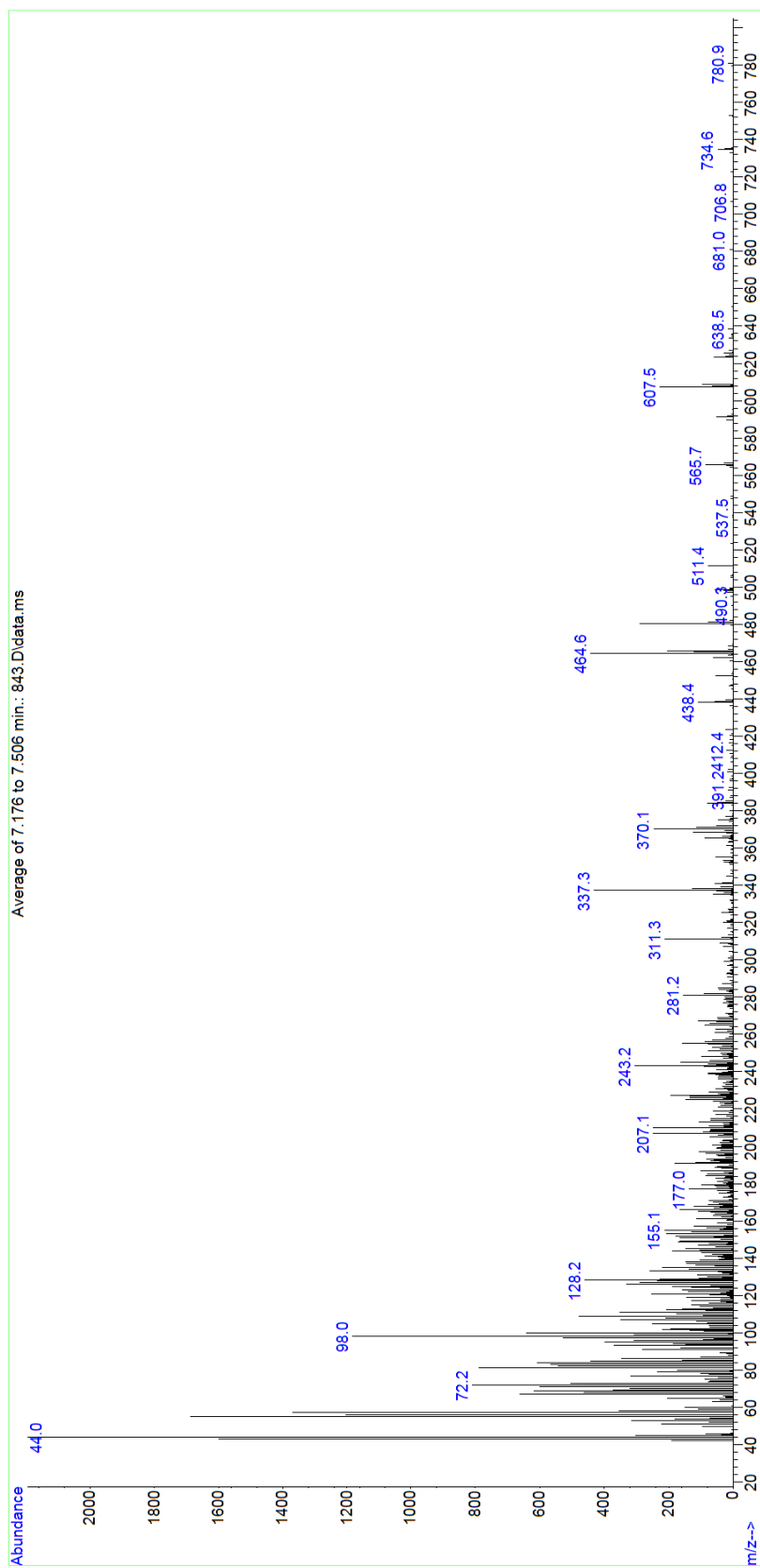


Figure 5. ^1H , ^{13}C HMBC NMR spectrum of compound **8**

Figure 6. EIMS spectrum of compound **8**



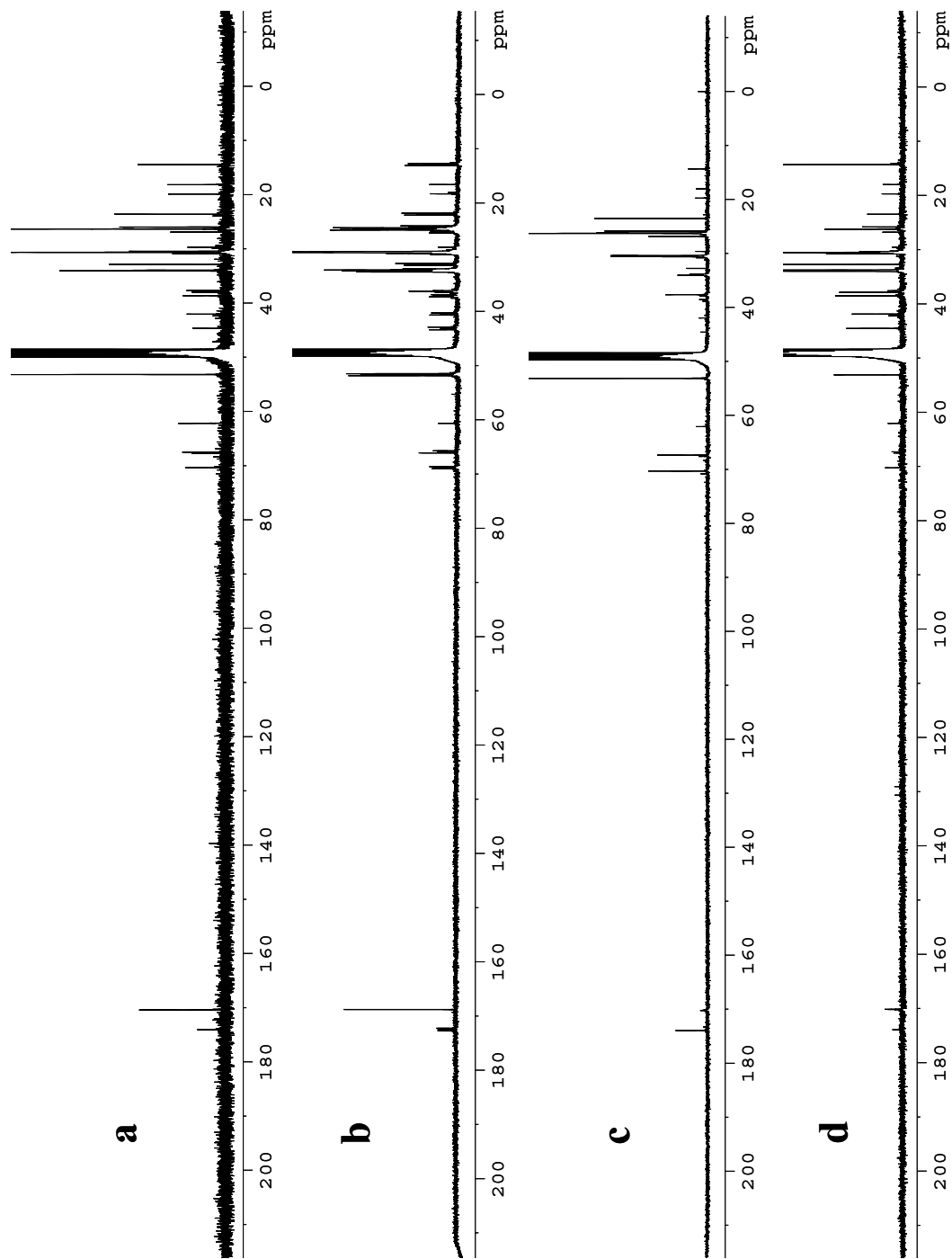


Figure 7. ^{13}C NMR spectra of (a) compound **8** and (b) $[1,2-^{13}\text{C}]$ acetate, (c) $[1-^{13}\text{C}]$ acetate and (d) $[2-^{13}\text{C}]$ acetate labeled compound **8**

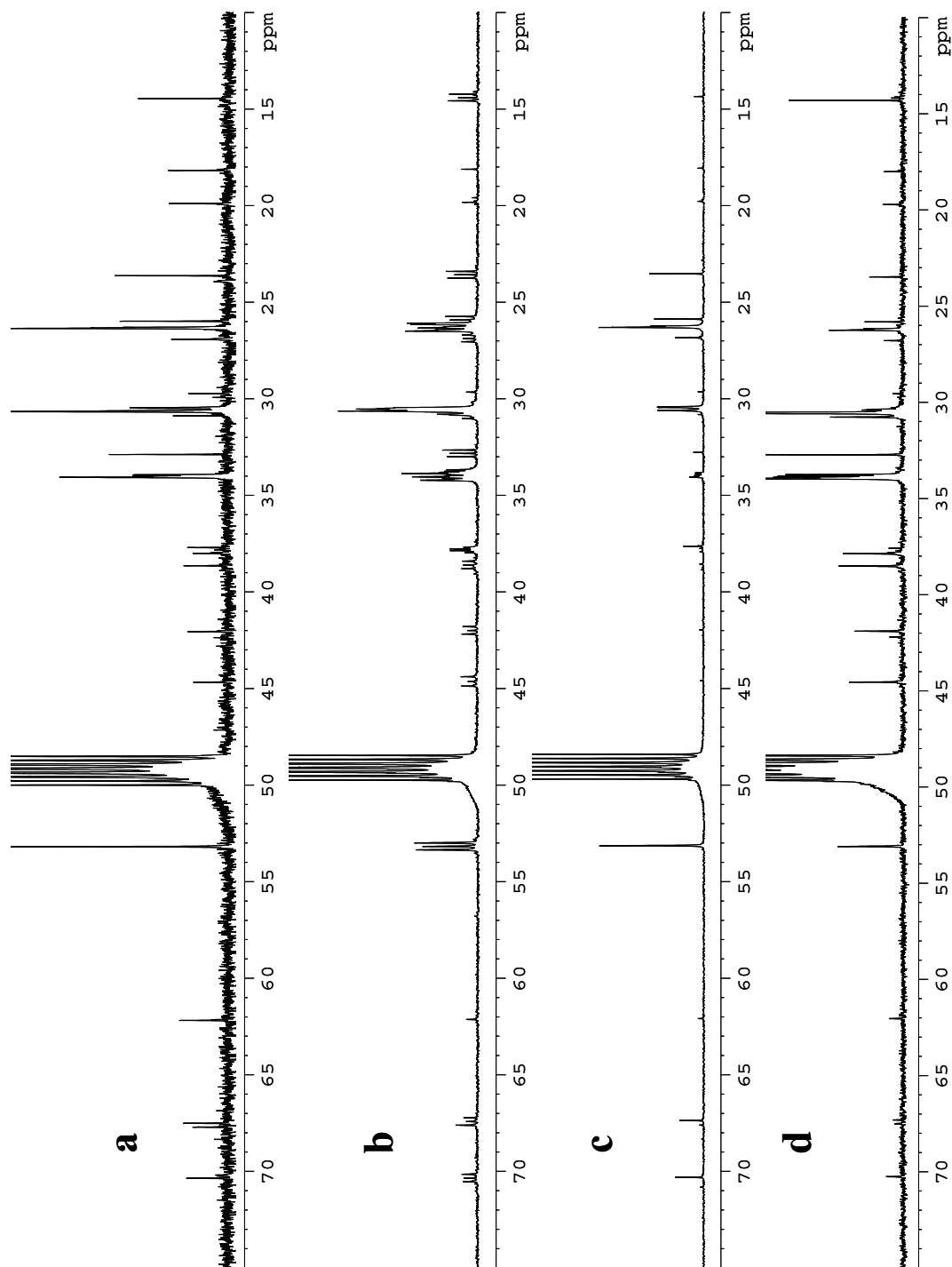


Figure 8. 10-75 ppm of Figure 8.

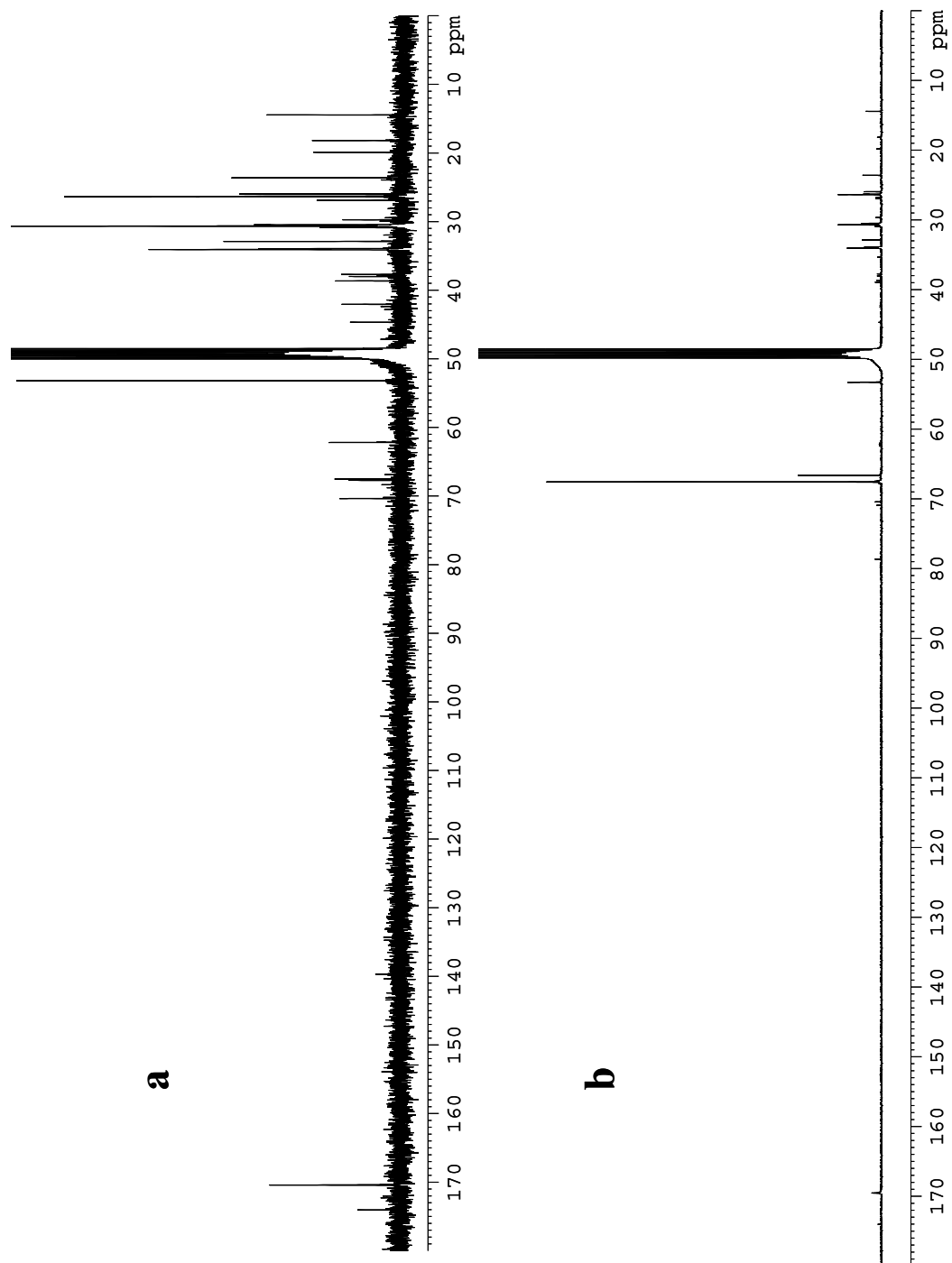


Figure 9. ^{13}C NMR spectra of (a) compound **8** and (b) $[1-^{13}\text{C}]$ -valine labeled compound **8**